Technical Notes

THE EFFECT OF PHYSICAL AND CHEMICAL RESTRAINT ON SELECTED RESPIRATORY PARAMETERS OF *MACACA MULATTA*[1]

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SUMMARY • A comparison of the respiration of Macaca mulatta restrained with strait jackets or injected intramuscularly with 1 mg/kg of phencyclidine hydrochloride revealed that the minute volume of the strait-jacketed animals was about 3 times that of relatively unrestrained animals when measurements were made 10, 20, and 30 minutes after treatment. The values obtained from the drug-treated animals were about 60% of those predicted by Guyton (3), and those obtained from strait-jacketed monkeys were about 200% of Guyton's predicted values. These differences were found to be statistically significant in every case. The 2 treatments also produced significantly different respiration rates (drug-treated monkeys about 67% of the strait-jacketed animals), heart rates (drug-treated rate about 84% of the jacketed monkeys), and tidal volumes (drug-treated about 52% of the jacketed animals' volumes).

The use of monkeys for the study of airborne disease usually requires some method of restraint to protect animals, personnel, and equipment. The administration of drugs such as pentobarbital sodium or phencyclidine hydrochloride (Sernylan®, Parke, Davis & Company, Detroit, Michigan) or the use of physical restraints such as strait jackets, taping of hands and feet, or caging are the procedures most commonly employed.

The effect of sedation or restraint on host respiration is important because aerosol dosage estimations are usually based on the animal's respiratory minute volume (4). Volumes in this laboratory are commonly computed from formulas published by Guyton (3) for a number of laboratory animals. Guyton's formula for the rhesus monkey is:

$$\text{min vol (ml/min)} = 2.35 \text{ (wt in gm)}^{3/4}$$

Body weight is the only variable considered, since no account is taken of the varying physiological states of the animal. Guyton made all of his measurements on what he called "normal" animals. "Resting" or "quiescent" would perhaps be more appropriate adjectives.

Changes in the respiratory measurements of laboratory animals as a result of various treatments have been reported by a number of workers. A good compilation of these data is found in the *Handbook of Respiration* (1). Unfortunately, little of this work has been done with monkeys, and this handbook lists only 3 references for the effects of drugs on these animals. In each case,

the drug employed, pentobarbital and thiopental (5) and phosgene (2), depressed the respiratory minute volume.

This report presents the data obtained from a series of experiments in which the respiratory measurements obtained from physically restrained animals (strait jacket) were compared with those obtained from drug-treated animals (phencyclidine) and with the minute volumes predicted by Guyton (3).

MATERIALS AND METHODS

Experimental animals—The *Macaca mulatta* (rhesus monkeys) used in this study weighed 2,000–3,600 gm. They were held in quarantine for 12 weeks before use. They were housed in wire-bar cages, fed monkey chow[3] twice daily, and had free access to water. The animals were fasted on the day of use.

Methods of Measurement—Respiratory minute volumes were determined by placing the monkey's head in a plastic mask that was connected to a dry gas meter. The gas meter was calibrated by comparing the meter reading with the volume displaced by a 50-ml syringe previously calibrated gravimetrically. A cyclic error was observed which at no time exceeded ± 10%. Appropriate check valves were included so that only expired air was measured. An attempt was made to keep hose connections short and the mask tight-fitting to minimize dead space. Respiration rates were determined by counting the pulsations of the pointer of the meter. Heart beats were counted with a stethoscope applied at the time of measurement.

EXPERIMENTAL PROCEDURES AND RESULTS

Restraint Procedures—The monkey was removed from its holding cage, placed in a plastic chair, and restrained by taping its hands and feet with masking tape. The mask which was connected to the meter was placed over the monkey's head and allowed to remain for 4 minutes before readings were taken. Respiration rate, minute volume, and heart rate were determined. Tidal volumes were calculated by dividing the minute volume by the respiration rate. Readings were taken at 0, 10, and 20 minutes with a 4-minute mask period preceding each. After each reading, the mask was removed, and the system was air-washed to remove condensed water vapor. After the first 3 readings, the animals were removed from the chair and either wrapped in the strait jacket or given an intramuscular injection of 1 mg of Sernylan per kg of body weight. Readings were then taken 10, 20, and 30 minutes after treatment. The effect of each treatment was determined on each of 20 animals in each of 3 experiments following a plan of treatment randomization.

Respirometric Findings—The respiratory minute volumes, respiration rates, heart rates, and tidal volumes of the animals restrained with strait jackets were found to be significantly different by statistical analysis from the corresponding values of the drug-treated monkeys (Table 1) regardless of the posttreatment

[3] Ralston Purina Company, St. Louis, Missouri.

TABLE 1

Respirometric values obtained with Sernylan®-treated and strait-jacketed monkeys

| Statistic | Parameter studied | Pretreatment | | | Results at indicated time (min) Posttreatment | | | | | | Guyton's min vol (ml/min) |
		0	10	20	10 Sernyl	10 Jacket	20 Sernyl	20 Jacket	30 Sernyl	30 Jacket	
Means*	Minute volume (ml/min)	2136	1606	1487	642	1842	515	1650	530	1587	901
	Respiration rate (breaths/min)	51.4	46.8	46.3	39.8	61.9	39.3	60.0	41.0	58.1	—
	Heart rate (beats/min)	242	242	237	203	243	198	235	196	234	—
	Tidal volume (ml/breath)	41.5	33.8	31.2	15.9	28.8	13.6	27.1	13.1	26.6	—
Coefficient of variation (%)	Minute volume	32.3	41.9	50.5	63.4	46.6	49.3	45.7	40.8	44.7	—
	Respiration rate	15.2	20.1	25.3	21.0	22.5	22.7	20.2	19.3	21.3	—
	Heart rate	12.8	14.8	15.5	27.9	13.8	20.5	15.8	23.9	18.7	—
	Tidal volume	28.7	28.3	31.2	48.1	33.3	52.3	36.7	42.0	31.6	—
95% confidence limits for minute volume	Upper limit	2315	1780	1682	747	2064	580	1845	586	1770	
	Lower limit	1957	1432	1294	537	1620	449	1455	474	1403	

* Means of determination on 60 monkeys.

period considered. Further, both treatments produced results significantly different (P ≤ 0.01) from those predicted by Guyton (3) at all observation periods. Finally, a marked effect of time was noted with every parameter (Table 1). Minute volumes declined markedly during the 20-minute pretreatment period as well as after treatment with phencyclidine. Restraint of the animals with a strait jacket produced an initial rise in minute volume, respiration, and heart rates. This rise was followed by a decline, but the magnitude was not as great as that seen in the animals prior to treatment.

DISCUSSION

The findings of this study indicate that the method and duration of restraint significantly affect the respiratory pattern of the rhesus monkeys. Of equal importance is the finding that, in every case, the minute volumes differed significantly from those predicted on the basis of Guyton's equation (3). Any calculation of minute volume based upon this formula would be in error by a significant amount, in many cases by several hundred percent. It is apparent, as Guyton states (3), that an extended rest period (considerably more than the 20 minutes in this study) would be required before the animals become sufficiently quiescent to permit the use of his predicted volume.

Perhaps the most important conclusion that can be drawn from this work is that accuracy in determining the respiratory minute volume of a monkey can best be achieved by measurement at the time of experimentation. A study is in progress to develop such a procedure. Alternatively, it may be possible to derive a mathematical formulation for the predication of minute volume that would take into account the physiological state of the animal in addition to its size (weight). This possibility is also under investigation.

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